Study: Kamel et al (2007) **Quality:** Moderate (7 pts)

F Kamel, CM Tanner, DM Umbach, JA Hoppin, MCR Alavanja, A Blair, K Comyns, SM Goldman, M Korell, JW Langston, GW Ross, DP Sandler; Pesticide Exposure and Self-reported Parkinson's Disease in the Agricultural Health Study. Am J Epidemiol 2007, 165 (4): 364-374.

STUDY SUMMARY

Study Overview

The Agricultural Health Study (AHS) cohort is used to assess the association of pesticide exposure and Parkinson's disease (PD). The AHS cohort was enrolled in 1993-1997, and follow-up interviews were conducted 1999-2003. Subjects were pesticide applicators and their spouses. At enrollment, subjects provided detailed information on lifetime pesticide use. At enrollment and follow-up, questionnaires elicited a response to whether the subject reported a physician diagnosed PD. There were 83 reported enrollment PD cases (prevalent cases) and 78 reported follow-up PD cases (incident cases). Incident PD was associated with cumulative days of pesticide use (p=0.009). Prevalent PD was not associated with overall pesticide use. The study suggests that exposure to certain pesticides may increase PD risk. Potential herbicides associated with PD are pendimethalin, paraquat, cyanazine, dicamba, trifuralin, 2,4,5-trichlorophenoxyacetic acid (2,4,5-T), and butylate, though only cyanazine attained statistical significance and only for prevalent PD. Potential other pesticides associated with PD are: the fumigants, carbon disulfide/carbon tetra-chloride, ethylene dibromide, and methyl bromide; the insecticides, lindane and phorate; and the fungicides, chlorothalonil and benomyl. Atrazine was among the herbicides found to be not statistically associated with PD.

Study Details

Study Participants. The AHS cohort was established in 1993–1997. At enrollment, 52,393 private applicators completed a self-administered enrollment questionnaire. Most (43,692; 83 percent) of the enrolled private applicators were married, and 32,345 (74 percent) of their spouses enrolled in the study by completing a spouse questionnaire. Hence there were 84,738 subjects at enrollment. The enrollment questionnaires elicited information on pesticide use, demographic factors, lifestyle, and medical history, including physician-diagnosed PD.

Five years after enrollment, 57,251 cohort members (68% of 84,738 enrollment cohort) completed a follow-up questionnaire. Information from the follow-up interviews used for analysis was self-reported physician-diagnosed PD, age at diagnosis, age at interview, and smoking status.

Exposure Measurement. The initial exposure measurement is 'ever use of any pesticide', where 82 percent of enrollment subjects indicated yes. The ever pesticide use was further broken down to mixing pesticides (categorized as no, <50% time, ≥50% time, and missing) and applying pesticides (also categorized as no, <50% time, ≥50% time, and missing). The ever pesticide use was further reported by duration (in years) and frequency (days per year) of use of any pesticide. Cumulative days of use of any pesticide was the product of duration and frequency, categorized by quartiles. Finally, the ever use of pesticide was further dichotomized by ever use of 50 commonly used pesticides. One the 50 was atrazine; simazine and propazine were not among the 50.

Additional exposure measurements are 'use of personal protective equipment', 'pesticide related medical care', and 'washing after a high personal exposure event'. Applicators provided information on use of types of personal protective equipment (e.g., chemical-resistant gloves), and from these responses 'use of personal protective equipment' was categorized as low protection, moderate protection, high protection, or missing. For applicators, information on pesticide-related medical attention was collected using the question, "As a result of using pesticides, how often have you seen a doctor or been hospitalized?" Responses were categorized as no, yes, or missing. Based on two questions, 'washing after a high personal exposure' was categorized as no event, washed within 1 hour, washed after 1 hour, or missing.

Outcome Ascertainment. At enrollment and follow-up, subjects were asked, "Has a doctor ever told you that you had been diagnosed with Parkinson's disease?'. There were 83 reported enrollment PD cases (prevalent cases). A small number of possible cases were excluded due to inconsistent responses (e.g., responding yes at enrollment and no at follow-up). There were 78 reported follow-up PD cases (incident cases). These incident cases were first diagnosed with PD after enrollment and before follow-up (by responding no or missing at enrollment and yes at follow-up).

Methods of Analysis. Logistic regression was used to evaluate the relation of either prevalent PD or incident PD to general pesticide variables. Models included adjustment for age at enrollment; state (Iowa or North Carolina); and type of participant (applicator or spouse).

Confounders Considered. Models included adjustment for age at enrollment; state (Iowa or North Carolina); and type of participant (applicator or spouse). Most applicators were men (>99 percent) and most spouses were women (96 percent), so no further adjustment was made for gender. Other confounders considered were race, education, and smoking.

Effect Measure and Point Estimates. There was a weak, not statistically significant, inverse association of prevalent PD with ever use of any pesticide (OR, 0.5; 95% CI, 0.2-1.1), with personally mixing pesticides ≥50% time (OR, 0.7; 95% CI, 0.3-1.5), and with personally applying pesticides ≥50% time (OR, 0.6; 95% CI, 0.3-1.2). In contrast, there was a weak positive, not statistically significant, association of incident PD with ever use of any pesticide (OR, 1.3; 95% CI, 0.5-3.3), with personally mixing pesticides ≥50% time (OR, 1.2; 95% CI, 0.5-2.7), and with personally applying pesticides ≥50% time (OR, 1.9; 95% CI, 0.7-4.7). Cumulative lifetime days of use was associated with a dose-related increase in incident PD ($p=0.009$), but not prevalent PD ($p=0.49$). The incident PD odds ratio of highest to lowest quartile on cumulative lifetime days was 2.3 (95% CI, 1.2-4.5).

Considering only pesticides for which there were four or more exposed cases, odds ratios for prevalent PD were elevated (≥1.4) for: the herbicides pendimethalin, paraquat, and cyanazine; and the fumigants carbon disulfide/carbon tetra-chloride and ethylene dibromide. Odds ratios for incident PD were elevated (≥1.4) for the herbicides dicamba, trifuralin, 2,4,5-trichlorophenoxyacetic acid (2,4,5-T), and butylate; the insecticides lindane and phorate; the fungicides chlorothalonil and benomyl; and the fumigant methyl bromide. Atrazine use was not statistically associated with prevalent PD (OR, 1.0; 95% CI, 0.5-1.9), nor with incident PD (OR, 1.1; 95% CI, 0.5-2.2).

Strength and Limitations Discussed in the Paper

A notable strength of the study is the use of AHS, a large sample from an agricultural population with many pesticide exposed subjects. Other strengths noted in the paper include its ability to distinguish between prevalent and incident cases, representation of diverse farming practices (Iowa v NC), and subjects providing detailed exposure data (including 50 specific pesticides).

At enrollment, subjects and prevalent PD cases with high pesticide exposure may have been less likely to enroll in the study than those with lower exposure, because PD or other effects of pesticide exposure led them to stop farming at an earlier age prior to possible enrollment. The questionnaires focused on lifetime pesticide use; where pesticide use may decrease over time due to earlier health effects. Among other limitations noted are the self-reporting of PD, self-reporting of pesticide usage, the challenge of considering concurrently multiple pesticides, potential bias due to only 68% subjects from enrollment completing follow-up, and lacking date of diagnosis for prevalent PD cases (hence unknown duration of PD at enrollment). Finally, though not explicitly acknowledged, the authors were aware that subset analyses (e.g., separate analyses by applicator and spouse or by smoking status) lacked statistical power.

EVALUATION

Study Design

This study design consists of two parts: the enrollment data forms a retrospective cohort substudy to investigate association of lifetime pesticide use with current PD status (i.e., prevalent PD). With the addition of follow-up interviews, the second part is a prospective cohort substudy to investigate association of lifetime pesticide use with onset of PD (i.e., incident PD). While the enrollment cohort is large (84,738 subjects), PD is a relatively rare disease (83 prevalent PD cases and 78 incident PD cases). Hence, the statistical power is low.

Statistical Analysis and Data-related Issues

Logistic regression is an adequate analytic methodology. The authors elected to use a two-stage hierarchical logistic regression to increase precision (Witte et al, 2000). Conceptually, this two-stage model makes sense, i.e., pesticides classified by functional groups (e.g., insecticides, herbicides, etc.) and by chemical groups (e.g., organophosphates, etc.) form a second stage hierarchy of specific pesticides. However, no results are shown to demonstrate any increased precision. Further, the resulting Bayesian analysis uses a prior residual variance of 0.35 as suggested by Witte et al. (2000). No justification for this selection is provided.

The categorization of all demographic variables (e.g., age as 12-50, 51-60, 61-70, 71-92) was found reasonable. The categorization of exposure variables (e.g., cumulative lifetime days of pesticide use as 0-64, 65-200, 201-396, ≥397) was also found reasonable. Although, in a supplemental regression model, cumulative days of pesticide use was used as a continuous covariate defined by the midpoints of the levels in the categorical variable. First, there appears to be no reason why the actual days could not have been used, and second, the midpoint of the interval ≥397 days is not clear.

The logistic regressions adjust for age, state, and type of participant (applicator/spouse). Other potential confounders (gender, smoking, race, education) were not used in the regresssions. Type of participant and gender were highly correlated, since applicators tended to be male, hence due to collinearity only type of participant was used as a covariate. Smoking tended to reduce the risk of PD (consistent with other publications), but was found to be not statistically significant. Unreported regressions which adjust for smoking did not change associations of PD with pesticide use.

Other unreported regressions (stratified analyses by type of applicator, stratified analyses by state, adjustment by a 'weighted' cumulative days of pesticide use, etc.) also did not substantively change any findings. However, due to the relatively small sizes of PD, there was little statistical power to detect differences in any of these subsets.

Table 2 (in the paper) summarizes the association of PD with symptoms of PD. While it is reassuring that PD cases (particularly prevalent PD cases) have a higher tendency to exhibit symptoms of PD, this analysis is not necessary to the goals of the study. Limited mortality data was available, but not statistically analyzed, but considering mortality data is not necessary to the goals of the study.

Each of the ever use of 50 commonly used pesticides was available to be used in regressions, though only pesticides for which there were at least four exposed cases were included in the models. Few statistically significant odds ratios were reported. However, no adjustment was attempted for multiple comparisons (likely no statistically significant associations with specific pesticides would be found had any multiple comparison adjustment been employed).

Logistic regression on the enrollment data (with prevalent PD as the outcome) is an adequate analysis. Logistic regression on the follow-up data (with incident PD as the outcome) is also an adequate analysis. Since the date of PD diagnosis between enrollment and follow-up is known, an alternative analysis on the follow-up data could have been a Poisson regression.

Biases

The logistic regression on enrollment data is subject to biases. As the authors note, farmers with PD due to high exposure may have stopped farming at an earlier age (and not been eligible for enrollment). Also, 25 subjects with possible prevalent PD were excluded from analysis; 12 subjects excluded due to conflicting responses on enrollment questionnaires, and 13 subjects excluded due to denial of PD on follow-up questionnaire.

The logistic regression on follow-up data is also subject to biases. Follow-up questionnaires were completed by 68% of subjects enrolled. Supplemental analysis showed follow-up respondents were similar at enrollment to follow-up non-respondents. Nonetheless, potential bias exists due to 32% non-respondents.

Collinearity among the 50 specific pesticides is likely (i.e., a farmer who has ever used alachlor has likely also ever used metolachlor), though no analysis is shown. Such collinearity usually results in wider confidence intervals, but may also bias odds ratio estimates.

The logistic regressions adjust for age, state, and type of participant (applicator/spouse). Age is a known risk factor for PD. State and type of participant are probably not risk factors, but may be surrogate covariates for risk factors. The authors note that type of participant and gender are highly correlated. Type of participant also may be correlated with other attributes of pesticide use (the applicator's contact with pesticides is fundamental different from the spouse's contact). State may be a surrogate for some unmeasured variables (e.g., weather, crops, etc.).

Finally, self-reporting pesticide use and PD diagnosis was noted by the authors as a limitation. The authors cite other studies that have found AHS self-reporting to be reliable. Hence the attenuation and bias of associations in this study due to self-reporting is likely to be minimal.

Alternative Reasons for Observed Results

The odds of PD in North Carolina were lower than in Iowa for both prevalent PD (OR, 0.6; 95% CI, 0.4-0.9), and incident PD (OR, 0.6; 95% CI, 0.4-1.0). In addition to state being a surrogate for some unmeasured risk factors, state may be correlated with attributes of pesticide use. If so, adjusting for state in the regressions may be masking or diminishing effects of pesticide use.

Similarly, the odds of PD for spouses were lower than for applicators for both prevalent PD (OR, 0.7; 95% CI, 0.4-1.1), and incident PD (OR, 0.6; 95% CI, 0.4-1.0). Logically, the applicator should be at higher risk. However, adjusting for type of participant in the regressions may be masking or diminishing effects of pesticide use.

The cumulative lifetime days of pesticide use seems likely to be correlated with age. Similarly, many pairs of ever use specific pesticides are likely correlated. The interpretation of regressions with collinear risk factors should be made cautiously.

Finally, the two-stage hierarchical model is based on an analysis in Witte et al (2000). The analysis in Witte et al considers the association of food intake and breast cancer. While it seems reasonable to make analogous assumptions for these two models, there are a myriad of potential reasonable assumptions that have gone untested in this study.

REFERENCES CITED

Witte JS, Greenland S, Kim LL, et al. Multilevel modeling in epidemiology with GLIMMIX. Epidemiology 2000, 11:684–8.